

Pringsheim to be due to a peculiar red-fluorescing pigment, usually masked by the yellow pigments, which he called *protochlorophyll*. Liro³ observed that absolutely dark-grown seedlings do not have the etiolin-spectrum of Pringsheim unless they are exposed to the light for a few seconds or minutes, and rightly concluded that etiolin is a mixture of the *protochlorophyll* of Monteverde and *chlorophyll*.

Liro made an extensive study of the formation of *chlorophyll* in the phanerogams and concluded that *protochlorophyll* is a postmortal decomposition product of a colorless organic substance which develops in the dark, and is designated, after Sachs,⁴ as *leucophyll*. According to Liro, *leucophyll* decomposes to form *protochlorophyll* in cells that are killed, and changes photochemically into *chlorophyll* upon exposure to light. *Protochlorophyll* may be observed in living leaves, but Liro assumed this to be due to occasional dead cells in which the *leucophyll* has undergone decomposition. In support of this view Liro reports that a layer of five or more living leaves of etiolated seedlings of *Avena sativa*, *Triticum sativum* or *Sinapis alba* was necessary, when examined spectroscopically, for the detection of the absorption band characteristic of *protochlorophyll*, but after the leaves were killed by dipping them in boiling water a single leaf or a layer of only two leaves was sufficient to give an evident *protochlorophyll* spectrum. It is assumed that in killing the cells the *protochlorophyll* is formed as a decomposition product of *leucophyll*. This conclusion is contradicted by practically all the experiments in Liro's paper, as he claims that *leucophyll* changes quantitatively into *chlorophyll* in etiolated leaves upon exposure to light even though the cells have been killed in any manner whatsoever, as by freezing, by immersing in boiling water, by exposing to ether vapors, by drying, by grinding to a paste in a mortar. *Leucophyll* has never been extracted from plants, though Liro tried many substances as solvents. His conclusion is that the *leucophyll* was either destroyed by or was insoluble in each of the substances tried.

The presence in plants of *leucophyll*, in the sense of Liro, is entirely hypothetical. That *protochlorophyll* is a decomposition product of some other organic substance seems highly improbable in the light of recent studies on the formation of *chlorophyll* made in the Botanical Institute of the University of Erlangen, Germany. It has been found that absolutely dark-grown seedlings of *Zea Mays* and *Avena sativa* contain only *protochlorophyll* when extracted

in the dark, *protochlorophyll* and *chlorophyll* when extracted in red light in the dark room, and only *chlorophyll* when extracted in diffuse daylight in the laboratory. The relation of *protochlorophyll* to *chlorophyll* is always quantitative, and as the amount of *protochlorophyll* decreases under the influence of light the amount of *chlorophyll* increases. *Protochlorophyll* is changed into *chlorophyll* by red light which has no photochemical effect on orthochromatic photographic emulsions. As found by Liro, red light is about twenty times as effective as blue light in converting *protochlorophyll* (*leucophyll*, according to Liro) into *chlorophyll*. In daylight *protochlorophyll* changes quantitatively into *chlorophyll* very rapidly. Etiolated seedlings of *Avena sativa*, which had been air-dried in the dark-room and stored in a covered box in the laboratory for approximately one year, were extracted in the light and were found to contain *protochlorophyll* and magnesium-free *protochlorophyll*, but no *chlorophyll*. *Protochlorophyll* in ether solution does not change to *chlorophyll* when exposed to light.

Dr. Noack, of the Botanical Institute of the University of Erlangen, has made preparations of pure *protochlorophyll*, and established its chemical relationship with a similar pigment which occurs in the gall of animals.

From the results of the studies briefly referred to above, *protochlorophyll* is not a decomposition product of some other organic substance, as *leucophyll*, but is a pigment which develops without the influence of light and changes photochemically into *chlorophyll* upon exposure to light. It is probable that this change occurs only in the presence of a specific enzyme.

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INTERNATIONAL COMMISSION ON ZOOLOGICAL NOMENCLATURE OPINIONS 98 TO 104

THE undersigned has the honor to invite the attention of the zoological profession to the fact that Opinions 98 to 104 have been published by the Smithsonian Institution.¹ The summaries read as follows:

Opinion 98. Rigidly construed, Brauer and Bergensstamm (1889 to 1894) did not fix the types for the older generic names, except in the cases where they distinctly state that the species mentioned is the type of the genus.

¹ *Smithsonian Miscellaneous Collections*, 73; No. 5, pp. 1-28.

³ *Annales Academiae Scientiarum Fennicae*, Ser. A, Tom. 1. 1909.

⁴ *Lotos*, 1859.

Opinion 99. *Entamoeba* 1895, with *blattae* as type by subsequent (1912) designation, is absolute synonym of *Endamoeba* Leidy, 1879a, p. 300, type *blattae*, and invalidates *Entamoeba* 1895, type by subsequent (1913) designation *hominis* = *coli*.

Opinion 100. Under Suspension of the Rules the genotype of *Spirifer* Sowerby, 1816, is fixed as *Anomia striata* Martin, and the genotype of *Syringothyris* Winchell, 1863, is fixed as *Syringothyris typa* Winchell (= *Spirifer carteri* Hall).

Opinion 101. The technical Latin designations used by Danilewsky, 1891, *Annales de l'Institut Pasteur*, Vol. 5 (12), pp. 758-782, are not in harmony with the International Rules of Zoological Nomenclature and are therefore not subject to citation or the Law of Priority on basis of said publication.

Opinion 102. A generic name (example *Proteocephalus*, 1858) is not invalidated by the earlier publication of the identical or a similar name of higher rank (example *Proteocephala*, 1828). If *Taenia ambigua* (tod. of *Proteocephalus*, 1858) is congeneric with *ocellata* (tsd. of *Ichthyotaenia*, 1894), *Ichthyotaenia* is a subjective synonym of *Proteocephalus*.

Opinion 103. The type of *Grus* Pallas, 1767, is *Ardea grus* Linn., 1758, by absolute tautonymy. *Grus* is hereby placed in the Official List of Generic Names.

Opinion 104. The following 57 generic names, with type species cited, are hereby placed in the Official List of Generic Names:

PROTOZOA: *Bursaria*, *Eimeria*, *Laverania*, *Plasmodium*, *Sarcocystis*. CESTODA: *Ligula*. NEMATODA: *Filaria*, *Heterodera*, *Rhabditis*, *Strongylus*, *Syngamus*. OLIGOCHAETA: *Enchytraeus*. HIRUDINEA: *Haemadipsa*, *Limnatis*. CRUSTACEA: *Armadillidium*, *Astacus*, *Cancer*, *Diaptomus*, *Gammarus*, *Homarus*, *Nephrops*, *Oniscus*, *Pandalus*, *Penaeus*, *Porcellio*. XIPHOSURA: *Limulus*. SCORPIONIDEA: *Scorpio*. ARANEAE seu ARANEIDA: *Avicularia*, *Dendryphantès*, *Dysdera*, *Latrodectus*, *Segestria*. ACARINA: *Cheyletus*, *Chorioptes*, *Demodex*, *Dermanyssus*, *Glyciphagus*, *Polydesmus*, *Psoroptes*, *Rhizoglyphus*, *Trombidium*. THYSANURA: *Lepisma*. COLLEMBOLA: *Podura*. ORTHOPTERA: *Blatta*, *Ectobius*, *Gryllus*, *Periplaneta*. ANOPLERA: *Pediculus*, *Phthirus*. HEMIPTERA: *Anthocoris*, *Nabis*, *Notonecta*, *Reduvius*, *Triatoma*. DERMAPTERA: *Forficula*. SUCTORIA s. SIPHONAPTERA s. APHANIPTERA: *Pulex*. MAMMALIA: *Cercoptihæcus*.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

THE SUBCUTANEOUS LYMPH SAC OF THE FROG AS A CULTURE CHAMBER

MUCH of the tedium involved in maintaining tissue cultures may be avoided by taking advantage of the

natural culture chamber afforded by the subcutaneous lymph sacs of the frog. The large ventral lymph sac is especially suitable for this purpose. A slit cut in the ventral skin in the region of the sternum makes it possible to slip excised bits of various frog tissues into the ventral lymph sac, where conditions are favorable for a continuation of living processes in the explanted tissue. The lymph of the living host serves as an aseptic and nutritive medium for the explant, and the explanted tissue may be left undisturbed until the conclusion of the experiment.

Lymph-sac cultures of integument may be maintained for two months or more, but the differentiated cells of liver and kidney undergo early disintegration. Explants of stomach wall and lung wall are more often successful in the lymph sac than those of liver or kidney but do not persist as long as do cells of integument.

In cases where small pieces of integument were inserted into the ventral lymph sac with their epidermis in contact with the subcutaneous surface of the skin of the host, sections taken from a series of such cultures show that the epidermal cells move out from the cut edges of an explant and migrate along the surface of a lymph coagulum which forms between the explant and the subcutaneous surface of the ventral skin of the host. At about forty-eight hours after operation, the migrating cells complete the formation of a vesicle, the wall of which consists, in part, of a newly formed epithelial layer and, in part, of the original explant. The tendency of pieces of integument to form vesicles when cultivated in the frog lymph sac was reported by Winkler (1910), who made no attempt to account for the phenomenon.

About thirty days after operation, sections show that growth of the dermal cells of the explant has begun. This dermal growth continues until the vesicle mentioned above becomes completely invested with dermis. Beginning about three weeks after operation, the subcutaneous blood vessels of the host skin produce branches which invade the newly formed dermis of the vesicle. In later stages up to fifty-five days, these invading blood vessels are filled with normal red blood corpuscles, which fact indicates that these vessels are active in connecting the vesicle with the cutaneous circulation of the host. How long such a parasitic existence will persist can be determined only by further experimentation.

Cultures of frog integument in the lymph sac are equally successful, whether the explanted tissues and the host are of the same or of different species of frog. At least this is true in cases where the two species used are *Rana pipiens* and *Rana clamitans*.

This method of tissue culture fails to afford opportunities for direct observations upon living cells, but